II. REMARKS

Before the amendments made herein, claims 97-101 and 123-129 were pending. Claims 123 to 126 have been canceled herein without prejudice, and claims 130 to 133 added. Accordingly, after the amendments made herein are entered, claims 97 to 101, 127 to 129 and 130 to 133 will be pending.

A. Regarding the amendments

Claims 130 to 133 have been added and are directed to heparanase proteins that are, respectively, at least 70%, 80%, 90% or 95% homologous to SEQ ID NO: 10. The claims are supported by the specification, for example, at page 59, lines 1-9.

Because all of the amendments made herein are fully supported by the specification, no issue of new matter arises.

B. Regarding the claim objections

Claims 100, 101, 127 and 128 are objected to for being dependent on rejected claim 97. Indeed; the Office Action Summary lists claims 100, 101, 127 and 128 as objected to but not rejected. However, page 7 of the Office Action lists claims 100 and 101 as rejected under 35 U.S.C. § 102(b). Applicants therefore request clarification. In any event, claim 128 has been amended so as to be independent.

C. Regarding enablement

Claims 97 to 99 and 123 to 126 are rejected under 35 U.S.C. § 112, first paragraph, as allegedly not enabled by the specification. Applicants respectfully traverse the rejection, noting that claims 123 to 126 have been canceled herein without prejudice.

1. The Office Action fails to make a single substantive point in comparing the subject claims to that of *Sun*.

The Office Action addresses the *Sun* case by merely stating that "For applicants convenience it is pointed out to applicants that the referred to claims from *Ex parte Sun* are drawn to a wee1 'polynucleotide' having at least 80% identity to the entire coding region of SEQ ID NO: 1 and this is a 403 amino acid encoding polynucleotide. The instant claims are drawn to a "preparation comprising a heparanase protein" said heparanase having at least 80% homology to SEQ ID NO: 10, wherein said heparanase is pure enough to elicit anti-heparanase antibodies."

The Action goes on to say that it has not presented an "exhaustive list" comparing each of the Wands factors for the instant claims and that of Ex parte Sun. With all due respect, Applicants do not see where the Action has at all compared the instant claims to that of *Sun* regarding any of the Wands factors. While the Action states one the subject claims and a claim of *Sun*, no comparative analysis is actually made by the Action and Applicants are missing the point here.

2. The Action's allegation that Applicants have not compared the subject case to *Sun* beyond the percent homology claimed is completely untrue.

The Action goes on to say that "applicants have neither pointed out similarities between the two decisions [*Sun* and the subject application] beyond that each encompasses a percent homology or identity and a protein or polynucleotide." Respectfully, Applicants assert that this allegation is absolutely not true.

a. The chemical nature of the compounds being claimed here and in *Sun* is each disclosed.

In the response to the Office Action mailed October 30, 2005, presented a detailed list comparing the facts of instant case to that of *Sun*. Specifically, on page 14 of *Sun*, the Board rejected the examiner's position of non-enablement. First, the Board pointed out that the chemical structure of the claimed sequence is disclosed.

Similarly, the chemical structure of SEQ ID NO:10 is disclosed in the subject specification.

b. How to screen for activity of the compounds being claimed here and in *Sun* is each disclosed.

Second, the Board noted that the specification provides an example for how to screen for WEE1 activity. Similarly, the subject specification discloses how to screen for heparanase activity. See, for example, Figures 9a and b and page 47, lines 14-21, which show testing for such activity by measuring the level of peak II HS degradation fragments. See also the Examples section.

As an aside, the Action continues to take the position that anything greater than 95% homology with SEQ ID NO:10 encompasses more variants than can be routinely screened with a reasonable expectation of success. If this is true, why did the Board in *Sun* find that the variants encompassed by 80% homology to the WEE1 protein can be routinely screened with a reasonable expectation of success? The Office Action continues to fail to address this point.

c. How to vary the claimed compound of the subject claims and in *Sun* is each disclosed. Indeed, the information provided in the subject application is far, far greater than that provided in *Sun*.

Finally, and most importantly, the Board in *Sun* found that the specification enabling by giving **the most general guidance about where to vary the protein**. Specifically, "most of the variations in the amino acid sequences of WEE1 are in the amino terminus, while the carboxy end of the genes are relatively conserved."

By contrast, Applicants have given **far more** detailed guidance in modifying the heparanase protein of the subject invention that the guidance cited by the Board in *Sun* regarding the WEE1 protein. Indeed, attached is a declaration by Dr. Iris Pecker, a named inventor of the subject invention, which summarizes previous data and provides additional data.

Specifically, there are three objective scientific reasons why the skilled artisan could have identified working embodiments that fall within the scope of claims 97, 98 and 99 with routine experimentation. First, the genetic drift or variation within the naturally occurring species of heparanase proteins, as shown in Figure 17 of the subject application and as described further below, provides a roadmap for variations including point mutations. Further variation can be gleaned from the secondary structure of the protein, as shown in Figure 19, which can provide for example the location of the proton donor. Second, the skilled artisan's knowledge of protein chemistry allows prediction of preserved biological properties so long as amino acid substitutions are conservative in their nature. Finally, the secondary structure of heparanase, as shown in Figure 19, would allow the skilled artisan to predict areas of non-criticality where even non-conservative substitutions may be introduced. Decl., para. 3.

i. Comparative analysis of the amino acid sequence of different species of heparanase.

The subject application, including Figure 17 provides ample guidance to the skilled artisan on how to make active heparanase variants that are at least 70% homologous with SEQ ID NO:10, and certainly variants that are at least 80% or 90% homologous with SEQ ID NO:10. For example, residues 85 to 106 of human heparananse (SEQ ID NO:10) are identical to the corresponding residues of the variant mouse and rat heparanases shown in Figure 17. It is important to note that these variants share less than 70% homology. By contrast, residues 23 to 36 have 11 residue differences. Similarly, comparing rat and human heparanase, residues 129 to 138, for example, have 8 differences among the 10 residues, with 9 of 10 differences among mouse and human at this region. With such guidance, the skilled artisan would know to not vary residues 85 to 106 and to vary one or more residues among residues 23 to 36 and/or 129 to 138, especially with a similar amino acid residue substitution (e.g., hydrophilic). The skilled artisan could even further use the guidance of the subject specification to replace one or more amino acid residues in

SEQ ID NO:10, especially in these highly variable regions, with those corresponding residues found in mouse or rat heparanase. Decl., para. 5.

Looking at heparanase protein more broadly, residues 49 to 109 make up 61 residues. Comparing mouse and human region at this region, there are only 10 of 61 changes. Comparing rat and human at this region, there are also only 10 of 61 changes. This is therefore a very conserved region, one that the skilled artisan would likely not vary, at least as a starting point, in trying to obtain additional heparanase homologs. Decl., para. 6.

The predictions discussed above have indeed become true. For example, the conserved region of residues 49 to 109 was confirmed to be the 8 kDa unit of active heparanase. By contrast, variable regions 23 to 36 and 129 to 138, discussed above, are not part of either the small or large units of mature heparanase. Decl., para. 7.

ii. The secondary structure of heparanase protein.

Moreover, Applicants point out for the first time in this case that Figure 19 of the subject application provides even further guidance. Figure 19 shows the secondary structure prediction for heparanase using computer assistance. The portions depicted as "H" are helical, and the portions depicted as "E" are extended beta strand structures. Decl., para. 8.

Still further, the glutamic acid residue of heparanase, predicted as the proton donor, is marked with an asterisk in Figure 19. Given the relative location of the proton donor and the predicted secondary structure of the protein, the glutamic acid residue that functions as the nucleophile is most likely at position 343 or 396 (see underlined residues in Figure 19, and page 105, lines 20-22 of the subject specification). Decl., para. 9.

In summary, given the wealth of information in the subject specification, particularly from figures 17 and 19, and the scientific technology available for exploiting this information, the skilled artisan can make heparanase variants at least

70% homologous with SEQ ID NO:10, and certainly at least 80% or 90% homologous with SEQ ID NO:10, without undue experimentation. The comparison of the data provided herein and the paucity of the data provided in *Sun* makes it clear that the instant claims are enabled.

Finally, Applicants point out that new claims 130 to 133 are directed to heparanase proteins with at least various percentage homologies with SEQ ID NO: 10, without "comprising" language. Applicants note that these claims more closely track the one the Action cited, apparently approvingly, from *Sun*.

In summary, the decision in *Sun* dictates that the subject claims are enabled by the specification. This is especially true where the claimed homology is 80% or even 90%. This is also especially true regarding new claims 130 to 133. Accordingly, Applicant respectfully requests that this rejection be withdrawn.

D. Regarding anticipation

Claims 97 to 101 and 123 to 126 are to be rejected under 35 U.S.C. § 102 as allegedly anticipated by Fuks et al. (U.S. Pat. No. 5,362,641; hereinafter "Fuks"). Applicants respectfully traverse the rejection, noting that claims 123 to 126 have been canceled herein without prejudice.

1. The difference between the subject claims and the preparation of Fuks is one of purity, not inherency.

The prior version of the subject claims recited that the claimed preparation of heparanase "be able" to elicit anti-heparanase antibodies. In response, the Examiner took the position that this "capability" to elicit such antibodies was inherent in Fuks.

Applicants then argued that if the subject claims were directed to 90% purity and Fuks could only get its preparation to 80% purity, would the Examiner argue that the preparation of Fuks had the potential to be 90% pure? In the Action mailed April 14, 2006, the Examiner disagreed with this analogy. Rather, he argued that an

appropriate analogy would be if the subject claims were directed to a preparation "capable of being purified to 90%" **not "purified to 90%."**

While respectfully disagreeing with the Examiner's interpretation of the phrase "being able," Applicants amended the claims to follow the Examiner's own logic. Therefore, claim 97 (and all claims dependent thereon) now recite "wherein said preparation **is pure enough** to elicit anti-heparanase antibodies."

To this, the current Office Action states that the heparanase protein taught by Fuks "is pure enough to elicit anti-heparanase antibodies even if the heparanase proteintaught by Fuks et al. was in a compostion that did not elicit anti-heparnase antibodies. The limitation/characteristic that the heparanase of the claimed invention "is pure enough to elicit anti-heparanase antibodies" is an inherent limitation/characteristic of the isolated heparanase taught by Fuks et al."

If the Action's current position is correct, then Applicants ask again – what is the difference between claiming a preparation that is 90% pure over art that teaches a preparation that is 80% pure? The subject claims are directed to a preparation that **is pure enough** to elicit anti-heparanase antibodies. The Action cannot possibly argue again that this means that the claimed preparation is merely "capable" of being purified so as to elicit such antibodies. **This is not what these words mean.** Rather, these words mean that the claimed preparation, as is, can actually elicit such antibodies. Moreover, even if there were any ambiguity in such words, which there is not, Applicants are stating that this is the meaning of the subject claims.

In summary, the claimed preparation, as is, is pure enough to elicit anti-heparanase antibodies. In contrast, the preparation of Fuks, as is, is not pure enough to do so. There is no inherency issue here. This is a purity issue.

2. Fuks' own inventor admits that its preparation was not pure enough to elicit anti-heparanase antibodies.

To complete the record, Applicants submit a declaration from Dr. Israel Vlodavsky, which was filed in the prosecution of a related European case. Dr. Vlodavsky, who is a named inventor of Fuks, admits that the preparation of Fuks could not elicit anti-heparanase antibodies but, rather, elicited antibodies to another protein.

3. The declaration of Dr. Iris Pecker, submitted herewith, gives two reasons why it would be unobvious to make the preparation of Fuks pure enough to elicit anti-heparanase antibodies.

Indeed, as described earlier in prosecution, Dr. Iris Pecker reviewed an analysis of the heparanase preparation taught by Fuks. This analysis shows that the heparanase of Fuks was inextricably mixed with a significant amount of at least six other proteins: PAI-1, Nexin-I, Vimentin, Grp94/endoplasmin, FLT receptor and Tryptase. Indeed, as declared by Dr. Vlodavsky and discussed above, the amount of these non-heparanase proteins present was so significant, that antibodies to one of these proteins (PAI-1) were elicited, while antibodies to heparanase could not be. Decl. of Dr. Iris Pecker, paras. 10 and 11.

There are two reasons why Fuks failed to achieve sufficient purity of their preparation so as to be able to elicit anti-heparanase antibodies. First, heparanase is "sticky." Second, the amount of heparanase found in cells is extremely low. For these two reasons, it would not have been obvious to purify the preparation of Fuks according to the methods of Fuks sufficiently so as to have a preparation that is pure enough to elicit anti-heparanase antibodies. Decl., para. 12.

a. Heparanase is "sticky."

One reason why it is so difficult and unobvious to purify the heparanase preparation of Fuks to the point where anti-heparanase antibodies can be elicited is because heparanase protein is very sticky to sugars. Consequently, heparanase is sticky to separation columns. This is known as non-specific affinity. Decl., para. 13.

Proteins can only be separated via one factor at a time, such as size, pH, etc. The stickier a protein is to a column, the less it will be able to be separated because the stickiness factor will always interfere with the primary separation factor that one is using. The result is a broader band in the separation column, which means the protein is still meshed with other proteins. Decl., para. 14.

Indeed, Fuks subjected their preparation to a further purification cycle according to the methods they taught. The results of their analysis of this preparation were provided to me and showed that this preparation contained many proteins, as much as nine of them in amounts sufficient for amino acid micro sequencing; in other words very high amounts. Decl., para. 15.

b. The amount of heparanase found in cells is extremely low.

The stickiness of heparanase is exacerbated by the fact that it is difficult and unobvious to acquire a sufficient amount of this protein. Indeed, the level of heparanase in most cell lines, including SKH is low. For example, heparanase is not detected in an extract of 10^6 SKH cells by anti-heparanase antibodies which can detect quantity in the nanogram range. Heparanase activity could not be detected in SKH cells using a DMB assay, which can also detect quantity even in the nanogram range. Furthermore, in the very sensitive ECM assay, which can detect a protein in the picogram range, more than 10^6 cells are required to detect heparanase activity. Decl., para. 16.

In fact, Fuks needed 20 liters of culture to start with to arrive at their preparation and still failed to achieve sufficient purity to elicit anti-heparanase antibodies. This is because the fraction of heparanase in the total cell extract is extremely small, as explained above, and the purification procedure to eliminate all other cellular proteins must include several chromatographic steps. When purifying a protein, which comprises a small fraction of the protein mixture, a significant loss of the protein of interest is expected in each chromatographic step. This is specifically true for heparanase, which is a non-covalently bound dimer that is easily disintegrated. The ECM assay, used to follow the enzyme during the purification

procedure is very sensitive and allows the detection of even picograms of protein. Following such a multi-step procedure one expects to yield an extremely small amount of protein, which would be too small even for evaluation of purity and obviously not sufficient to elicit anti-heparanase antibodies. Decl., para. 17.

Using Fuks' procedures, nine contaminants could be identified. Although one could continue and employ the additional column suggested by Fuks, which would decrease the amount of contaminants, it would also decrease the amount of heparanase to the point where it could not elicit anti-heparanase antibodies. Moreover, the additional column step would still not separate all proteins which were co-eluted up to this point. Decl., para. 18.

In stark contrast to the method taught by Fuks, the methods of the subject invention were able to avoid all of the difficulties discussed above and therefore result is a preparation that is pure enough to elicit anti-heparanase antibodies. Specifically, using the heparanase gene, heparanase can be over-expressed. This yields an extract which is a-priori enriched with heparanase. The loss of heparanase during chromatography is therefore insignificant. Moreover, the availability of the sequence allows prediction of the protein pI, and other physicochemical characteristics that enable the design of a rational and efficient purification procedure. Using such an expression system as the source of protein, due to the high expected yield of heparanase, contaminants that may be co-eluted with heparanase would comprise a negligible proportion of the purified protein. Decl., paras. 19 and 20.

In summary, claims 97 to 101 are clearly directed to a preparation of heparanase that, as is, can elicit anti-heparanse antibodies. By contrast, by an inventor of Fuks' own admission, the preparation of Fuks, as is, can not elicit such antibodies. This is no different than a difference in purity. Therefore, Fuks does not anticipate subject claims 97 to 101.

Moreover, claims 97 to 101 are unobvious over Fuks. It would not have been obvious to use the methods of Fuks to purify their preparation so that it could elicit anti-heparanase antibodies because of the sticky nature of the protein and because it is present is such extremely low amounts in cells. Indeed, Applicants were able to

obtain their preparation, which is pure enough to elicit anti-heparanase antibodies, by a completely different way than Fuks – namely, by sequencing and over-expressing heparanase. For all these reasons, Applicants respectfully request that this rejection be withdrawn.

III. CONCLUSION

All of the issues raised in the Office Action have been addressed and are believed to have been overcome. Accordingly, it is respectfully submitted that all the claims under examination in the subject application are allowable. Therefore Applicants respectfully request a Notice of Allowance to this effect.

Respectfully submitted,

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Date: February 1, 2007

Enclosed:

Request for Continued Examination (RCE);
Declaration of Dr. Iris Pecker; and
Declaration of Israel Vlodavsky.